Parkinson's disease: molecular risk factors

Grünblatt, Edna

Abstract: Parkinson's disease (PD) is a progressive neurodegenerative disorder second only to Alzheimer's disease. Diagnosis remains clinical, based on phenotypic patterns. In the last decade many attempts to develop early differential pre-clinical markers have been reported. In this presentation, the molecular risk factors that may link between the etiopathogenesis leading to PD and peripheral markers will be discussed. Genetic variation known to be involved in familial forms of PD will be shown to be linked to sporadic cases, as for example leucine-rich repeat kinase 2 (LRRK2) that was found to regulate microRNA-mediated translation regulation. In addition postmortem microarray findings of transcription alterations will be compared to the peripheral findings of mRNA profiles. Molecular processes involved in ubiquitination and proteasome, autophagy, mitochondrial dysfunction and the nicotinic and adenosine A2 protection will be discussed. The question of what time-point should be used measuring the different markers and the course of the disease considered, and the future possibilities in exploring these techniques will be debated.

DOI: https://doi.org/10.1016/S1353-8020(11)70016-5

Parkinson's disease: Molecular risk factors

Edna Grünblatt

Hospital of Child and Adolescent Psychiatry, University of Zurich, Neumuensterallee 9, 8032 Zurich, Switzerland

Page no. 14 Abstract: 161 words Text: 1733 Words Table: 1

References: 19

Key Words: Parkinson's disease, transcription, RNA, diagnose, marker, blood, postmortem

PL02 Hirotaro Narabayashi Symposium: From Pathology to Diagnosis

Chairpersons: Shengdi Chen *China* and Ronald F. Pfeiffer *USA*

✉ All correspondence to PD Dr. Edna Grünblatt:

Hospital of Child and Adolescent Psychiatry, University of Zurich, Neumünsterallee 9, CH-8008 Zurich, Switzerland

Tel: +41-44-578 60 71

Fax: +41-44-578 60 81

E-mail: edna.gruenblatt@kjpdzh.ch

Abstract

Parkinson's disease (PD) is a progressive neurodegenerative disorder second only to Alzheimer's disease. Diagnosis remains clinical, based on phenotypic patterns. In the last decade many attempts to develop early differential pre-clinical markers have been reported. In this presentation, the molecular risk factors that may link between the etiopathogenesis leading to PD and peripheral markers will be discussed. Genetic variation known to be involved in familial forms of PD will be shown to be linked to sporadic cases, as for example leucine-rich repeat kinase 2 (LRRK2) that was found to regulate microRNA-mediated translation regulation. In addition postmortem microarray findings of transcription alterations will be compared to the peripheral findings of mRNA profiles. Molecular processes involved in ubiquitination and proteasome, autophagy, mitochondrial dysfunction and the nicotinic and adenosine A2 protection will be discussed. The question of what time-point should be used measuring the different markers and the course of the disease considered, and the future possibilities in exploring these techniques will be debated.

Introduction

Parkinson's disease (PD) is a progressive neurodegenerative disorder second only to Alzheimer's disease. Since the fact that diagnosis remains clinical based on the presence of a combination of cardinal signs, including rest tremor, bradykinesia, rigidity, and loss of postural reflexes, the diagnosis will most often be made after more than 70-80% of the dopaminergic neurons have degenerated [1]. There is wide agreement that a preclinical approach detecting at-risk PD subjects would enable earlier preventive therapy that might slow down or even stop the neurodegeneration. In this review the molecular risk factors will be described, beginning with familial genetic findings and continuing with transcription profile studies in the central nervous system (CNS)that have been studied postmortem to the new findings of transcription factors, alterations found in peripheral blood samples, which link some again to familial genetic forms.

Since the discovery of the first disease-causing mutation in the α-synuclein gene (*SNCA*/ PARK1/4), a number of genes and loci have been implicated in PD denoted as PARK1 to 14. Those genes harboring mutations that cause late-onset dominantly inherited PD include *SNCA* and leucine-rich repeat kinase 2 (*LRRK2*/PARK8), whereas early-onset recessive parkinsonism is associated with homozygous and compound heterozygous mutations in *parkin* (*PRKN*/PARK2), Pten-induced kinase 1 (PINK1/PARK6) and Oncogene *DJ-1* (DJ-1/PARK7) [2]. Despite their importance in identifying key molecules in the pathogenesis of PD, these genetic mutations remain relatively rare (less than 5%). On the other hand, association studies in sporadic PD have found that similar genes such as SNCA polymorphism, as well as microtubule-associated protein tau/saitohin (*MAPT*/*STH*), glucoberebrosidase (*GBA*) and *LRRK2* are associated with the risk of PD, but as the highest odds ratio after meta-analysis reaches 3.4 it must still be validated in larger and diverse population [3].

As an additional approach to finding new candidate molecules involved in the etiopathogenesis of PD, many gene expression studies have been conducted on postmortem brain tissue, especially on the substantia nigra (SN), the affected brain region (for review see Table 1).

Studying the various reports and different brain regions reveals the involvement of protein metabolism and handling (e.g. aggregation and ubiquitination processes, proteasome and vesicular trafficking), the energy pathway including the mitochondria, the response to stress (e.g. heat shock proteins and their chaperones), the autophagy processes, inflammation, adenosine receptors (caffeine) as well as oxidative metabolism (Table 1). In addition, in many of the postmortem studies found a link between familial to sporadic PD, with the finding of alterations in the transcription of *SNCA* [6, 8, 9, 12], which may point to a common neurodegeneration mechanism in both cases. Similarly, *UCHL-1* (PARK5) and *DJ-1* (PARK7) mRNA were found to alter in sporadic PD [4-6, 10].

As the availability of postmortem brain tissue is limited, as well as the need for early diagnosis research has been directed to the periphery, with the aim of finding similar alterations there as in the CNS. Indeed, such alterations have been confirmed [6]. As in the postmortem brain studies, many of the molecular pathways discovered in the first approach have also been found in the periphery (Table 1). *TRIM24* mRNA was found to alter its expression in peripheral blood both in sporadic PD as well as in carriers of the *PARK2* or *LRRK2* mutations [19]. *TRIM24* is involved in the transcription control of some nuclear receptors, but was also found to be involved in the control of apoptosis and autophagia mechanisms. Therefore it could be hypothesized that this marker may point to the common mechanism involving autophagy and oxidative stress leading to apoptosis. In PD carriers of the *LRRK2* mutation gene expression profiles in blood samples revealed the involvement of the

ubiquitin-proteasome system [18]. Again, also in this study one can conclude similar molecular pathways of the protein metabolism to be involved both in sporadic and familial PD. In a familial PD study with duplications of the *SNCA* gene it was shown that this duplication directly influences the expression of the *SNCA* mRNA in blood [18]. This finding enhances the notion of the involvement of *SNCA* in the etiopathology of PD.

From the current findings, it could be concluded that peripheral molecular markers can mirror the disease processes and may be even detect progression of the disease. Of course, the path is still long, in order to reach the best set of markers that will best provide early differential diagnosis for PD. But such future perspectives seem to be nearing. For such an aim, there is a need for a longitudinal large scale international study, in which at-risk subjects will be followed up for their progression to PD phenotypes while in parallel different markers will be collected, as for example peripheral blood samples (for DNA/ genotypes, RNA/ transcriptomics and proteins/ proteomics), imaging data as well as later on for conformational diagnosis, postmortem brain tissue should be collected when possible. Of course, since the data produced from such large gene wide analysis is huge and quite complicated there is need for interdisciplinary work field including molecular biologists, imaging experts, clinicians and bioinformatics specialist who can work this mass of information into comprehensive data. Such an approach could unravel new and common mechanism both involved in familial and sporadic PD that may lead to some new models for the neuropathophysiology causing this neurodegeneration. Still the main question of the time point, when one should start the diagnosis for PD, is not yet answered. The main reason to this is the fact that therapy should be available in order to slow-down or stop the progress of the disease. But also ethical aspects are involved.

References (Max 20)

[1] Wu Y, Le W and Jankovic J Preclinical biomarkers of Parkinson disease Arch Neurol 2011; 68**:** 22-30.

[2] Zheng B, Liao Z, Locascio JJ, Lesniak KA, Roderick SS, Watt ML, et al. PGC-1alpha, a potential therapeutic target for early intervention in Parkinson's disease Sci Transl Med 2010; 2**:** 52ra73.

[3] Lill C, Roehr J, McQueen M, Kavvoura F, Bagade S, Schjeide B, et al. The PDGene Database 5 September 2011 http://www.pdgene.org/

[4] Mandel S, Amit T, Kalfon L and Youdim MB Applying transcriptomic and proteomic knowledge to Parkinson's disease drug discovery Exp. Opin. Drug Discov. 2007; 2**:** 1225-40.

[5] Papapetropoulos S and McCorquodale D Gene-expression profiling in Parkinson’s disease: discovery of valid biomarkers, molecular targets and biochemical pathways Future Neurology 2007; 2**:** 29-38.

[6] Grünblatt E, Zehetmayer S, Jacob CP, Muller T, Jost WH and Riederer P Pilot study: peripheral biomarkers for diagnosing sporadic Parkinson's disease J Neural Transm 2010.

[7] Lu L, Neff F, Alvarez-Fischer D, Henze C, Xie Y, Oertel WH, et al. Gene expression profiling of Lewy body-bearing neurons in Parkinson's disease Exp Neurol 2005; 195**:** 27-39.

[8] Fuchs J, Tichopad A, Golub Y, Munz M, Schweitzer KJ, Wolf B, et al. Genetic variability in the SNCA gene influences alpha-synuclein levels in the blood and brain FASEB J 2008; 22**:** 1327-34.

[9] Shehadeh L, Mitsi G, Adi N, Bishopric N and Papapetropoulos S Expression of Lewy body protein septin 4 in postmortem brain of Parkinson's disease and control subjects Mov Disord 2009; 24**:** 204-10.

[10] Kumaran R, Vandrovcova J, Luk C, Sharma S, Renton A, Wood NW, et al. Differential DJ-1 gene expression in Parkinson's disease Neurobiol Dis 2009; 36**:** 393-400.

[11] Varani K, Vincenzi F, Tosi A, Gessi S, Casetta I, Granieri G, et al. A2A adenosine receptor over expression and functionality, as well as TNF-alpha levels, correlate with motor symptoms in Parkinson's disease FASEB J 2010; 24**:** 587-98.

[12] Shehadeh LA, Yu K, Wang L, Guevara A, Singer C, Vance J, et al. SRRM2, a potential blood biomarker revealing high alternative splicing in Parkinson's disease PLoS One 2010; 5**:** e9104.

[13] Lanoue AC, Dumitriu A, Myers RH and Soghomonian JJ Decreased glutamic acid decarboxylase mRNA expression in prefrontal cortex in Parkinson's disease Exp Neurol 2010; 226**:** 207-17.

[14] Nagai Y, Ueno S, Saeki Y, Soga F, Hirano M and Yanagihara T Decrease of the D3 dopamine receptor mRNA expression in lymphocytes from patients with Parkinson's disease Neurology 1996; 46**:** 791-5.

[15] Kawaguchi N, Yamada T, Takahashi M and Hattori T Expression of MxA mRNA in peripheral blood mononuclear cells in Parkinson's disease Parkinsonism Relat Disord 1999; 5**:** 43-7.

[16] Soreq L, Israel Z, Bergman H and Soreq H Advanced microarray analysis highlights modified neuro-immune signaling in nucleated blood cells from Parkinson's disease patients J Neuroimmunol 2008; 201-202**:** 227-36.

[17] Reale M, Iarlori C, Thomas A, Gambi D, Perfetti B, Di Nicola M, et al. Peripheral cytokines profile in Parkinson's disease Brain Behav Immun 2009; 23**:** 55-63.

[18] Mutez E, Lepretre F, Le Rhun E, Larvor L, Duflot A, Mouroux V, et al. SNCA locus duplication carriers: from genetics to Parkinson disease phenotypes Hum Mutat 2011; 32**:** E2079-90.

[19] Aguiar P and Severino P Biomarkers in Parkinson disease: global gene expression analysis in peripheral blood from patients with and without mutations in PARK2 and PARK8 einstein 2010; 8**:** 293-7.

Table 1: Summary of transcription (RNA) profiles in postmortem brain tissue and peripheral blood samples from Parkinson's disease subjects (modified from [4, 5])

Samples origin	**Region /cell type**	**Method of measurements**	**Genes found**	**Familial / Sporadic PD / HC**	**Reference / the research group**
CNS	SN	Affymetrix HG-Focus array	PSMA2-5 HIP2 SKP1A GBE1 UQCRC2 ALDH1A1 LAMB2 DNAJB5 (Hsp40) HSPA8	Sporadic	[6] Grünblatt and colleagues 2004
	SN, Putamen, BA9	Affymetrix U133A GeneChip	NEFL HSPB1 FGF13 NSF SNAP25 SYT1	Sporadic	[5] Zhang and colleagues 2005
	SN	Affymetrix U133A GeneChip	HSPA1A-B SYT1 UBE1	Sporadic	[5] Hauser and colleagues 2005
	LB-containing DA neurons vs. non-LB-containing DA neurons	RNA fingerprinting	STCH (Hsp70) USP8 ANP32B		[7]
	SN medial / lateral	Affymetrix U133A & B GeneChip	NEFL HIP2 HSPB1 UCHL-1 GBE1 ALDH1A1 FGF13 NSF SNAP25	Sporadic	[4, 5] Moran and colleagues 2006 Duke and colleagues 2006
	SN, Striatum	CodeLink Human	UCHL-1	Sporadic	[5]

Samples origin	**Region /cell type**	**Method of measurements**	**Genes found**	**Familial / Sporadic PD / HC**	**Reference / the research group**
		UniSet 20 K bioarray	FGF13 NSF SYN1-2 SYT1 STX1A		Miller and colleagues 2006
	Putamen, occipital cortex, cerebellar hemispheres	Affymetrix U133A GeneChip	NPY HTR2C SYT1	Sporadic	[5] Vogt and colleagues 2006
	SN, vetral tegmental area, perirhinal corte (BA35), insular cortex, amygdale, nucleus basalis, caudate, putamen, nucleus accumbens, golobus pallidus, mediodorsal thalamus, pulvinar, subthalamic nucleus, dorsal nucleus of the vagus nerve, cerebellar hemisphere, anterior cerebellar vermis, dorsal raphe, locus ceruleus,	Affymetrix Human Genome U133 Plus 2.0 GeneChip	MRPS6 STIP	Sporadic	[5] Papapetropoulos and colleagues 2006

Samples origin	Region /cell type	Method of measurements	Genes found	Familial / Sporadic PD / HC	Reference / the research group
	hypothalamus, hippocampus, reticular formation				
	Midbrain-SN	qRT-PCR	SNCA	Sporadic	[6] Chiba-Falek and colleagues 2006
	SN, amygdala	qRT-PCR	SNCA	Sporadic	[6] Papapetropoulos and colleagues 2007
	SN, cerebellum	qRT-PCR, Genotyping	SNCA	Sporadic	[8]
	NM-containing DA SN neurons	qRT-PCR	SNCA TH ENO2	Sporadic	[6] Gründemann and colleagues 2008
	NM-containing DA SN neurons	Affymetrix U133A GeneChip	SNCA UCHL-1 HIP2 PINK1 SOD1 HSPA8 UBE1-3 PSMB4-5/C3/D4 SYN1 NSF SYT1	Sporadic	[6] Simunovic and colleagues 2009
	SN, amygdala	qRT-PCR	SEPT4 SNCA	Sporadic	[9]
	Putamen, frontal cortex, parietal cortex, amygdala, cerebellum	qRT-PCR	DJ-1	Sporadic	[10]
	Putamen	qRT-PCR	$A_{2A}AR$	Sporadic	[11]

Samples origin	**Region /cell type**	**Method of measurements**	**Genes found**	**Familial / Sporadic PD / HC**	**Reference / the research group**
	SN, amygdala	qRT-PCR	SEPT4 SNCA	Sporadic	[12]
	BA9	In-situ hybridization; One-Color Agilent 60-mer Whole Human Genome Microarray	GAD76	Sporadic	[13]
	SN	genome-wide meta-analysis	PGC-1alpha	Sporadic	[2]
Peripheral blood	lymphocytes	Semi-quantitative PCR	D3R	Sporadic	[14]
	PBMC	Semi-quantitative PCR	MxA	Sporadic	[15]
	Whole blood	Affymetrix U133A GeneChip	ST13 HIP2 CLTB	Sporadic	[2] Scherzer and colleagues 2007
	Whole blood	Affymetrix U133A GeneChip; CodeLink; Stanford	SNCA ALAS2 FECH BLVRB	HC	[2] Scherzer and colleagues 2008
	Whole blood	Affymetrix U133A GeneChip	SNCA DBH ST13	Sporadic	[16]
	PBMC	Semi-quantitative PCR	MIP-1 alpha MCP-1 IL-8 IL-1 beta IFN gama TNF alpha	Sporadic	[17]
	Whole blood	qRT-PCR	PSMA2-5 LAMB2 ALDH1A HIP2 HIST1H3E	Sporadic	[6]
	lymphocytes	qRT-PCR	$A_{2A}AR$	Sporadic	[11]

Samples origin	Region /cell type	Method of measurements	Genes found	Familial / Sporadic PD / HC	Reference / the research group
	Whole blood	Affymetrix Exon_ST1	SRRM2 long isoform	Sporadic	[12]
	PBMC	Agilent one-colour whole human genome 44K microarray; Genotyping	ALAS2, ARG1 DUSP10 ERAF IFNG LTF SELENBP1 UTG2B17	PD carriers of LRRK2 mutation	[18] Mutez and colleagues 2010
	Whole blood	Affymetrix Human Gene 1.0 st.	TRIM24	Sporadic; PARK2, PARK8	[19]
	leukocytes	qRT-PCR; comparative genomic hybridization; one-color whole human genome 44K microarrays	SNCA FAM13A	Familial with SNCA duplication	[18]

Annotation: $A_{2A}AR$, adenosine 2A receptor; ALAS2, 5-aminolevulinate synthase 2; ALDH1A1, aldehyde dehydrogenase 1 family member A1; ANP32B, acidic (leucine-rich) nuclear phosphoprotein 32; ARG1, arginase, liver; BA9, Brodmann's Area 9; BLVRB, biliverdin reductase B; CLTB, clathrin, light chain B; CNS, central nervous system; D3R, dopamine receptor D3; DA, dopaminergic; DBH, dopamine beta-hydroxylase; DJ-1, PARK7; DNAJB5, DnaJ (Hsp40) homologue, subfamily B, member 5; DUSP10, dual specificity phosphatase 10; ENO2, neuron-specific enolase; ERAF, Erythroid associated factor; FAM13A, family with sequence similarity 13, member A; FECH, ferochelatase; FGF13, fibroblast growth factor 13; GBE1, Glucan (1,4-α-), branching enzyme 1 (glucagen branching enzyme, Andersen disease, glycogen storage disease type IV); HC, healthy controls; GAD67, glutamic acid decarboxylase 67kDa isoform; HIP2, huntigntin interacting protein 2 (PARK5); HIST1H3E, histone cluster-1 H3e; HSPA1A-B, heat shock protein A1A-B; HSPA8, Heat shock protein A8; HSPB1, heat shock 27 kDa protein 1; HTR2C, 5-hyproxytryptamine (serotonin) receptor 2C; IFNG, Interferon gamma; IR, insulin receptor, LAMB2, laminin, beta-2 (laminin S); LB, Lewy Bodies; LTF,

Lactotransferrin; MRPS6, mitochondrial ribosomal protein S6; NEFL, neurofilament, light polypeptide; NM, neuromelanin; NPY, neuropeptide Y; NSF, N-ethyl maleimide-sensitive factor; PBMC, peripheral blood momonuclear cells; PD, Parkinson's disease; PGC-1a, proliferator–activated receptor gama coactivator-1alpha PINK1, PTEN induced putative kinase 1 (PARK6); PSMA2-5, proteasome subunit alpha type 2-5; PSMB4-5/C3/D4, proteasome (prosome, macropain) subunit beta type 4-5/ 26S subunit ATPase3/ non-ATPase 4; qRT-PCR, quantitative RT-PCR; SELENBP1, Selenium binding protein 1; SEPT4, septin 4; SKP1A, S-phase kinase-associated protein 1A (p19A); SN, substantia nigra; SNAP25, synaptosomal-assocoated protein; SNCA, Alpha-synuclein (PARK1); SOD1, superoxide disomutase 1; SRRM2, serine/arginine repetitive matrix 2; ST13, suppression of tumorigenicity 13; STCH, stress 70 protein chaperone; STIP1, stress-induced phosphoprotein 1 (Hsp 70/Hsp90-organizing protein); STX1A, syntaxin 1A; SYN1-2. synapsin 1-2; SYT1, synaptotagmin 1, TH, tyrosine hydroxylase; TRIM24, tripartite motif containing 24; UBE1, ubiquitin-activating enzyme E1 UCHL-1, ubiquitin C-terminal hydrolase (PARK5); UQCRC2, Ubiquinol-cytochrome c reductase core protein II; USP8, ubiquitin specific protease 8; UTG2B17, UDP glucuronosyl transferase 2, B17;